

IN THE CLAIMS

1. (Previously presented) A method of inhibiting mucus secretion by a mucus-secreting cell, comprising administering to said cell a mucus-inhibitory amount of an active fragment of a MARCKS protein that inhibits MARCKS protein-related mucus secretion, wherein said fragment has a sequence comprising from 10 to 50 contiguous amino acids from SEQ ID NO: 3, or an amino acid sequence comprising an allelic variant, wherein said variant comprises deletions or replacements in said sequence, wherein said replacements are selected from the group consisting of:

- Val or Ser substituted for Ala;
- Ala, Leu, Met, or Ile, substituted for Val;
- Ala, Val or Ile substituted for Leu;
- Pro or Cys substituted for Gly;
- Gly, Cys, Ser, or Met substituted for Pro;
- Gly, Pro, Ser, or Met substituted for Cys;
- Pro or Cys substituted for Met;
- Phe or Gln substituted for His;
- His, Tyr, or Trp substituted for Phe;
- His, Phe or Trp substituted for Tyr;
- Phe or Tyr substituted for Trp;
- Gln or Ser substituted for Asn;
- His, Lys, Glu, Asn, or Ser substituted for Gln;
- Gln, Thr, Pro, Cys or Ala substituted for Ser;
- Gln or Ser substituted for Thr;
- Gln or Arg substituted for Lys;
- Lys, Asp or Glu substituted for Arg;
- Lys, Arg, or Glu substituted for Asp;
- Arg or Asp substituted for Glu; and
- combinations of any of the foregoing; and

whereby mucus secretion by said cell is reduced compared to that which would occur in the absence of said active fragment.

2-5. (Canceled).

6. (Previously Presented) A method according to claim 1 wherein said method further comprises administering a compound selected from the group consisting of okadaic acid, calphostin C, Rp-8-Br-cGMP and LY83583.

7. (Original) A method according to claim 1 wherein said mucus-secreting cell is an epithelial cell contained within airway mucous membranes or gastrointestinal mucous membranes.

8. (Original) A method according to claim 1, wherein said compound is administered to the airways of a mammalian subject.

9. (Original) A method according to claim 1, wherein said compound is administered to the gastrointestinal tract of a mammalian subject.

10. (Original) A method according to claim 1 wherein said compound is administered by inhalation.

11. (Previously presented) A method of inhibiting mucus secretion by a mucus-secreting cell, comprising administering to said cell a peptide inhibitor of MARCKS-related mucus secretion, wherein said peptide inhibitor is selected from the group consisting of

(a) SEQ ID NO: 1;

(b) an active fragment of a MARCKS protein comprising from 10 to 50 contiguous amino acids from SEQ ID NO:3; and

(c) an amino acid sequence comprising an allelic variant, wherein said variant comprises deletions or replacements in the sequence of (a) or (b) above, wherein said replacements are selected from the group consisting of:

Val or Ser substituted for Ala;

Ala, Leu, Met, or Ile, substituted for Val;

Ala, Val or Ile substituted for Leu;

Pro or Cys substituted for Gly;

Gly, Cys, Ser, or Met substituted for Pro;
Gly, Pro, Ser, or Met substituted for Cys;
Pro or Cys substituted for Met;
Phe or Gln substituted for His;
His, Tyr, or Trp substituted for Phe;
His, Phe or Trp substituted for Tyr;
Phe or Tyr substituted for Trp;
Gln or Ser substituted for Asn;
His, Lys, Glu, Asn, or Ser substituted for Gln;
Gln, Thr, Pro, Cys or Ala substituted for Ser;
Gln or Ser substituted for Thr;
Gln or Arg substituted for Lys;
Lys, Asp or Glu substituted for Arg
Lys, Arg, or Glu substituted for Asp;
Arg or Asp substituted for Glu; and
combinations of any of the foregoing;

such that mucus secretion by said cell is inhibited compared to that which would occur in the absence of said peptide.

12-13. (Canceled).

14. (Original) A method according to claim 11 wherein said mucus-secreting cell is an epithelial cell contained within airway mucous membranes or gastrointestinal mucous membranes.

15. (Original) A method according to claim 11, wherein said peptide is administered to the airways of a mammalian subject.

16. (Original) A method according to claim 11, wherein said peptide is administered to the gastrointestinal tract of a mammalian subject.

17. (Original) A method according to claim 15 wherein said peptide is administered by inhalation.

18. (Previously presented) A method of inhibiting mucus secretion in the airways of a subject in need of such treatment, comprising administering to the airways of said subject a mucus-inhibiting amount of a compound that inhibits the MARCKS-related release of mucin, wherein said compound is selected from the group consisting of

(a) SEQ ID NO: 1;

(b) an active fragment of a MARCKS protein comprising from 10 to 50 contiguous amino acids from SEQ ID NO:3;

(c) an amino acid sequence comprising an allelic variant, wherein said variant comprises deletions or replacements in the sequence of (a) or (b) above, wherein said replacements are selected from the group consisting of:

Val or Ser substituted for Ala;

Ala, Leu, Met, or Ile, substituted for Val;

Ala, Val or Ile substituted for Leu;

Pro or Cys substituted for Gly;

Gly, Cys, Ser, or Met substituted for Pro;

Gly, Pro, Ser, or Met substituted for Cys;

Pro or Cys substituted for Met;

Phe or Gln substituted for His;

His, Tyr, or Trp substituted for Phe;

His, Phe or Trp substituted for Tyr;

Phe or Tyr substituted for Trp;

Gln or Ser substituted for Asn;

His, Lys, Glu, Asn, or Ser substituted for Gln;

Gln, Thr, Pro, Cys or Ala substituted for Ser;

Gln or Ser substituted for Thr;

Gln or Arg substituted for Lys;

Lys, Asp or Glu substituted for Arg

Lys, Arg, or Glu substituted for Asp;

Arg or Asp substituted for Glu; and

combinations of any of the foregoing; and
whereby mucus secretion in the airways of the subject is reduced compared to that which would occur in the absence of said treatment.

19. (Original) A method according to claim 18 wherein said subject is a mammalian subject suffering from a condition selected from the group consisting of bronchitis, asthma, cystic fibrosis, chronic obstructive pulmonary disease, emphysema, pneumonia, influenza, rhinitis and the common cold.

20-23. (Canceled).

24. (Previously Presented) A method according to claim 18, further comprising administering a compound selected from the group consisting of okadaic acid, calphostin C, Rp-8-Br-cGMP) and LY83583.

25. (Original) A method according to claim 18 wherein said compound is administered by inhalation.

26-38 (Cancelled).

39. (Previously presented) A pharmaceutical formulation comprising a mucus-inhibiting peptide fragment of MARCKS, wherein said mucus-inhibiting peptide fragment is selected from the group consisting of

(a) SEQ ID NO: 1;

(b) an active fragment of a MARCKS protein comprising from 10 to 50 contiguous amino acids from SEQ ID NO:3; and

(c) an amino acid sequence comprising an allelic variant, wherein said variant comprises deletions or replacements in the sequence of (a) or (b) above, wherein said replacements are selected from the group consisting of:

Val or Ser substituted for Ala;

Ala, Leu, Met, or Ile, substituted for Val;

Ala, Val or Ile substituted for Leu;

Pro or Cys substituted for Gly;
Gly, Cys, Ser, or Met substituted for Pro;
Gly, Pro, Ser, or Met substituted for Cys;
Pro or Cys substituted for Met;
Phe or Gln substituted for His;
His, Tyr, or Trp substituted for Phe;
His, Phe or Trp substituted for Tyr;
Phe or Tyr substituted for Trp;
Gln or Ser substituted for Asn;
His, Lys, Glu, Asn, or Ser substituted for Gln;
Gln, Thr, Pro, Cys or Ala substituted for Ser;
Gln or Ser substituted for Thr;
Gln or Arg substituted for Lys;
Lys, Asp or Glu substituted for Arg
Lys, Arg, or Glu substituted for Asp;
Arg or Asp substituted for Glu; and
combinations of any of the foregoing; and
a pharmaceutically acceptable carrier.

40-41. (Canceled).

42. (Original) A pharmaceutical formulation according to claim 39 where said composition is aerosolized.

43. (Original) A pharmaceutical formulation according to claim 39 where said peptides are contained within liposomes.

44-47. (Canceled).

48. (Previously presented) A method of inhibiting mucus secretion by a mucus-secreting cell, comprising administering to said cell a mucus-inhibitory amount of a compound, wherein said compound is a peptide having an amino acid sequence that

comprises from 10 to 50 contiguous amino acids from an N-terminal sequence of a MARCKS protein or an amino acid sequence comprising an allelic variant, wherein said variant comprises deletions or replacements in said sequence that binds to a target site selected from:

- (a) mucin granule membranes at the site bound by MARCKS protein; and
- (b) MARCKS protein at the mucin granule binding site; and

wherein said replacement is selected from the group consisting of:

Val or Ser substituted for Ala;
Ala, Leu, Met, or Ile, substituted for Val;
Ala, Val or Ile substituted for Leu;
Pro or Cys substituted for Gly;
Gly, Cys, Ser, or Met substituted for Pro;
Gly, Pro, Ser, or Met substituted for Cys;
Pro or Cys substituted for Met;
Phe or Gln substituted for His;
His, Tyr, or Trp substituted for Phe;
His, Phe or Trp substituted for Tyr;
Phe or Tyr substituted for Trp;
Gln or Ser substituted for Asn;
His, Lys, Glu, Asn, or Ser substituted for Gln;
Gln, Thr, Pro, Cys or Ala substituted for Ser;
Gln or Ser substituted for Thr;
Gln or Arg substituted for Lys;
Lys, Asp or Glu substituted for Arg;
Lys, Arg, or Glu substituted for Asp;
Arg or Asp substituted for Glu; and
combinations of any of the foregoing;

wherein the amount of mucus secreted by said cell is reduced compared to that which would occur in the absence of said compound.

50. (Currently Amended) A method according to claim 49 48 ~~where~~ wherein said peptide is myristoylated.

51-66 (Cancelled).

67. (Previously Presented) The method of Claim 1, wherein the N-terminal region of the active fragment of a MARCKS protein is myristoylated.

68. (Previously Presented) The method of Claim 1, wherein said active fragment of a MARCKS protein corresponds to the myristoylated N-terminus of MARCKS.

69. (Previously Presented) The method of Claim 1, wherein said active fragment of a MARCKS protein comprises SEQ ID NO: 1.

70. (Previously Presented) The method of Claim 18, wherein the N-terminal region of the active fragment of a MARCKS protein is myristoylated.

71. (Previously Presented) The method of Claim 18, wherein said active fragment of a MARCKS protein corresponds to the myristoylated N-terminus of MARCKS.

72. (Previously Presented) The method of Claim 39, wherein the N-terminal region of the active fragment of a MARCKS protein is myristoylated.

73. (Previously Presented) The method of Claim 39, wherein said active fragment of a MARCKS protein corresponds to the myristoylated N-terminus of MARCKS.

74. (Previously Presented) The method of Claim 48, wherein the N-terminal region of the active fragment of a MARCKS protein is myristoylated.

75. (Previously Presented) The method of Claim 48, wherein said active fragment of a MARCKS protein corresponds to the myristoylated N-terminus of MARCKS.

In re: Li et al.

Serial No.: 09/914,020

Filed: December 31, 2001

Page 10 of 11

76. (Previously Presented) A method of inhibiting mucus secretion by a mucus-secreting cell, comprising administering to said cell a peptide inhibitor of MARCKS-related mucus secretion, wherein said peptide inhibitor comprises SEQ ID NO: 1.